

K062546

SUMMARY OF SAFETY AND EFFECTIVENESS

IDENTIFICATION INFORMATION

FEB 14 2007

SUBMITTERS INFORMATION

This summary of 510(k) safety and effectiveness is being submitted in accordance with the requirements of SMDA 1990 and 21 CFR 807.20.

SUBMITTER'S NAME AND ADDRESS: Meridian Bioscience, Inc.
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CONTACT PERSON: Susan Rolih
Official correspondent

DATE SUMMARY PREPARED: August 29, 2006

TRADE NAME: ImmunoCard STAT! EHEC

COMMON NAME: *E coli* toxins detection test

CLASSIFICATION NAME: *Escherichia coli* serological reagents

REGULATION: 866.3255

INTENDED USES: ImmunoCard STAT! EHEC is an immunochromatographic rapid test for the qualitative detection of Shiga toxins 1 and 2 (also called Verotoxins) produced by *E. coli* in cultures derived from clinical stool specimens. ImmunoCard STAT! EHEC is used in conjunction with the patient's clinical symptoms and other laboratory tests to aid in the diagnosis of diseases caused by enterohemorrhagic *E. coli* (EHEC) infections.

PREDICATE DEVICE: Premier EHEC (K953362)

BACKGROUND: Among the *E. coli* human pathogens, Shiga toxin-producing strains of *E. coli* have gained in importance in recent years. The group of EHEC, with their highly pathogenic serovars O157:H7, 026, 0103, 0111, 0145, and other strains are of particular concern. Production of Shiga toxins is the most common criteria for the detection of this group of bacteria. Shiga toxins can be classified into two main categories: Shiga toxin 1 (ST1) and Shiga toxin 2 (ST2). EHEC strains may produce ST1 or ST2 only or both ST1 and ST2 simultaneously. EHEC are capable of initiating life-threatening illnesses, particularly in young children, the elderly or patients with immune deficiency. The main sources of infection are contaminated, raw or insufficiently heated foods of animal origin, eg, meat and dairy products. The reservoir for EHEC is the feces of cattle, sheep and goats. These microorganisms can enter food during the processing of meat and dairy products if hygienic conditions are inadequate. The incidence of food infection caused by Shiga toxin-producing *E. coli* demands reliable and rapid methods of detection. In addition to traditional culture methods, immunological techniques are becoming more useful due to their improved specificity and sensitivity. ImmunoCard STAT! EHEC is an immunological screening test based on the lateral flow principle.

This product was previously cleared to market as Duopath Verotoxin GLISA (K031367) to identify Shiga toxin 1 or Shiga toxin 2-producing strains of *E coli* isolated from colonies derived from human stools cultured on Sorbitol MacConkey agar plates. This application adds a new intended use to the device (use with broth cultures inoculated with human stool) and changes the name of the product.

Type of test

Qualitative, rapid, single use, immunochromatographic assay

Specimen type

Human stool samples inoculated into enrichment media (GN or Mac broth) (This application.)

Conditions for use

ImmunoCard STAT! EHEC is designed for use by laboratory professionals under normal environmental conditions. The assay, which is stored at 2-8 C when not in use, is brought to room temperature (20-25 C) before use. Normal laboratory lighting, humidity and temperature do not affect the performance of the assay.

Contraindications

There are no contraindications associated with the use of this product.

Special instrument requirements

No instruments are used with this device.

Combination with other medical devices

No other medical devices are used in combination with this device.

Table 1. Comparison of ImmunoCard STAT! EHEC to the predicate device and its prior design.

Characteristics	<i>ImmunoCard STAT! EHEC</i>	<i>Premier EHEC</i> (predicate)	<i>DuoPath Verotoxin GLISA</i> (predicate)
Device Type			
Technology	Single use, rapid, lateral flow immunoassay	Microwell-based enzyme-linked immunoassay	Single use, rapid, lateral flow immunoassay
In vitro diagnostic device	Yes	Yes	Yes
Control	Includes external control reagent	Includes external control reagent	No control reagent included
Calibrator	No	No	No
Assay Features			
Human factors	No special equipment	EIA-related equipment	No special equipment
Sterile device	No	No	No
Mechanical safety	Not applicable	Not applicable	Not applicable
Environmental safety	Normal medical waste	Normal medical waste	Normal medical waste
Chemical hazards	None	None	None
Radiation safety	Not applicable	Not applicable	Not applicable
Intended Use			
Detection of Shiga toxins 1 and 2	Yes	Yes	Yes
Differentiation between Shiga toxins 1 and 2	Yes	No	Yes
Screening test	Yes	Yes	Yes
Diagnostic test	No	No	No
Identification test	Yes	No	Yes
Monitoring therapy	No	No	No

This Table is continued on the next page.

Table 1 continued.

Acceptable Samples		<i>ImmunoCard STAT! EHEC</i>	<i>Premier EHEC</i> (predicate)	<i>DuoPath Verotoxin GL/SA</i>
Stool broth culture	Yes		Yes	No
Stool agar culture	Yes		No	Yes
Direct stool	No		Yes	No
Reagents/Components Provided				
Test Medium	Test Device with nitrocellulose strip	Antibody-coated microwell		Test Device with nitrocellulose strip
Conjugate Reagent	In Test Device	Stand alone reagent		In Test Device
Sample Diluent/Negative Control (external)	Yes	Yes		Yes
Substrate Reagent	No	Yes		No
Stop Solution	No	Yes		No
Internal positive run control	Yes	No		Yes
Internal negative run control	Yes	No		Yes
External positive control	Yes	Yes		No
Source of Antigens/Antibodies				
Capture ST1 antibodies	Murine monoclonal	Murine monoclonal		Murine monoclonal
Capture ST2 antibodies	Murine monoclonal	Murine monoclonal		Murine monoclonal
Detector ST1 Antibodies	Murine monoclonal	Rabbit polyclonal		Murine monoclonal
Detector ST2 Antibodies	Murine monoclonal	Rabbit polyclonal		Murine monoclonal
Positive Control	Inactivated toxin	Inactivated toxin		None
Comparison of assay steps*				
Equipment Required	None	EIA-related		None
Level of skill required	Moderately complex	Moderately complex		Moderately complex
Assay steps	5	15		5
End point	Pink-red band	Yellow color		Pink-red band
Interpretation of test result	Pos = color band, Neg = no color	OD ≥ 0.150 (dual wavelength)	Pos = color band, Neg = no color	Pos = color band, Neg = no color

This Table is continued on the next page.

Table 1 continued.

Performance Characteristics	<i>ImmunoCard STAT! EHEC</i>	<i>Premier EHEC (predicate)</i>	<i>Duopath Verotoxin GLISA</i>
Clinical Sensitivity	90.3%	100%	Not calculated
Clinical Specificity	100%	97.9%	Not calculated
Precision/Reproducibility (intra-assay)	100%	100%	100%
Linearity/reportable range	N/A	N/A	N/A
Analytical limit of detection/sensitivity	ST1 = 25 ug/mL ST2 = 25 ug/mL	ST1 = 7 pg/well ST2 = 15 pg/well	ST1 and ST2 = one colony per enrichment broth
Assay cutoff	N/A	OD 0.150 (dual wavelength)	N/A
Indeterminant range	N/A	N/A	N/A

DEVICE DESCRIPTION AND TECHNOLOGICAL PRINCIPLES

Construct

Each ImmunoCard STAT! EHEC test kit contains three reagents/components.

1. Test Devices, in individual foil pouches with desiccants.
2. Sample Diluent (Negative Control) in a plastic dropper vial
3. Positive Control in a plastic dropper vial

Calibrators

No calibrators are needed for this device.

Controls

This assay includes an internal control line that is used to demonstrate that the broth samples have been applied, that it has flowed correctly, and that the conjugated detector antibodies are active at the time of use. A colorless to faint pink background around the Toxin 1 and 2 Test Lines serves as a negative control and indicates the reagents were performing correctly at the time of testing.

The Positive Control external control reagent is used with the Sample Diluent/Negative Control when external control testing is warranted. The reagents also serve as indicators that the test was performed correctly, that reagents were active and specific at the time of use, and that the Test Device membrane supports proper sample flow.

Failure of the internal and external controls to produce the expected results suggests the test was not performed correctly.

Technological principles

ImmunoCard STAT! EHEC is an immunochromatographic rapid test utilizing monoclonal antibodies labeled with red-colored gold particles. The test device has a circular sample port and an oval-shaped test (Toxin 1, Toxin 2) and control (Control) window.

1. The sample is applied to the chromatography paper via the circular sample port (Sample).
2. The sample is absorbed through the pad to the reaction zone containing colloidal, gold-labeled antibodies specific to Shiga toxins.
3. Any Shiga toxin (ST1 and ST2) antigen present complexes with the gold-labeled antibody and migrates through the pad until it encounters the binding zones in the test (Toxin 1, Toxin 2) area.
4. The binding zones (Toxin 1 and Toxin 2) contain another anti-ST1 or -ST2 antibody, which immobilizes any Shiga toxin-antibody complex present. Due to the gold labeling, a distinct red line is then formed.
5. The remainder of the sample continues to migrate to another binding reagent zone within the control zone, and also forms a further distinct red line (positive control). Regardless of whether any Shiga toxin is present or not, a distinct red line should always be formed in the control zone and confirms that the test is working correctly.

CLINICAL TRIAL DATA

Performance evaluation

ImmunoCard STAT! EHEC is a modification of Duopath Verotoxin GLISA (K031367), cleared to market to detect EHEC in stool samples that had been cultured on Sorbitol MacConkey plates. This premarket submission describes the evaluations performed with clinical samples to validate the product's new intended use with broth cultures inoculated with human stool.

This study was conducted with samples tested fresh or following frozen storage. Samples were collected in the United States, Canada and Argentina. A total of 360 stool samples were evaluated in either GN, MacConkey (Mac) broths or both. 340 of these samples produced growth in GN broth, while 344 produced growth in Mac broth. Table 2 identifies the ages of the patients from whom samples were collected during the study. Table 3 provides the gender of each patient. Table 4 summarizes the sensitivity and specificity obtained with GN and Mac stool broth samples. Samples producing discrepant results between Premier EHEC and ImmunoCard

Table 3 Classification of patients from whom samples were collected based on gender.

	Specimen Type		
GN Samples	Male	Female	Not defined
Clinical site 3			
Total tested	37	51	213
Mean positive reaction strength	2.0	3.1	3.5
Positive reaction range	0.5-8.0	0.5-6.0	0.5-8.0
Clinical site 4			
Total tested	14	25	0
Mean positive reaction strength	1.0	All Negative	N/A
Positive reaction range	0.5-5.0	All Negative	N/A
Clinical site Totals			
Total tested	51	76	213
Mean positive reaction strength	1.9	3.1	3.5
Positive reaction range	0.5-5.0	0.5-6.0	0.5-8.0
	Specimen Type		
MAC Samples	Male	Female	Not defined
Clinical site 3			
Total tested	40	52	213
Mean positive reaction strength	2.8	3.3	3.3
Positive reaction range	0.5-7.0	0.5-7.0	0.5-6.0
Clinical site 4			
Total tested	14	25	0
Mean positive reaction strength	3.0	All Negative	N/A
Positive reaction range	3.0	All Negative	N/A
Clinical site Totals			
Total tested	54	77	213
Mean positive reaction strength	2.9	3.3	3.3
Positive reaction range	0.5-7.0	0.5-7.0	0.5-6.0

STAT! EHEC were further analyzed using cytotoxin assay (Table 5). These samples generally produced weak reactions (less than 0.300) in Premier EHEC (Table 6). Some of the samples grew in MAC broth but not GN broth.

Patient exclusion criteria

There are no patient exclusion criteria associated with ImmunoCard STAT! EHEC.

Influence of other disease states

There is no influence by other disease states on test results.

Table 2. Categories of patients by age from whom samples were collected for clinical studies

Patient Age and Sample Storage	GN Samples	birth to 1 month	>1 month to 2 years	>2 years to 12 years	>12 years to 21 years	>21 years	Not Defined
Clinical site 3							
Total tested	1	4	12	10	62	213	
Mean positive reaction strength	All Negative	All Negative	3.6	2.6	2.4	3.5	
Positive reaction range	All Negative	All Negative	1.0-5.0	0.5 - 6.0	0.5 - 6.0	0.5 - 8.0	
Clinical site 4							
Total tested	N/A	1	N/A	2	35	0	
Mean positive reaction strength	N/A	All Negative	N/A	All Negative	1.0	N/A	
Positive reaction range	N/A	All Negative	N/A	All Negative	1.0	N/A	
Clinical site Totals							
Total tested	1	5	12	12	97	213	
Mean positive reaction strength	All Negative	All Negative	3.6	2.6	2.3	All Negative	
Positive reaction range	All Negative	All Negative	1.0-5.0	0.5 - 6.0	0.5 - 6.0	All Negative	

Patient Age and Sample Storage	MAC Samples	birth to 1 month	>1 month to 2 years	>2 years to 12 years	>12 years to 21 years	>21 years	Not Defined
Clinical site 3							
Total tested	1	5	13	10	63	213	
Mean positive reaction strength	All Negative	All Negative	3.9	2.6	3.1	3.3	
Positive reaction range	All Negative	All Negative	0.5 - 6.0	0.5 - 7.0	0.5 - 7.0	0.5 - 6.0	
Clinical site 4							
Total tested	N/A	1	N/A	2	36	0	
Mean positive reaction strength	N/A	All Negative	N/A	All Negative	3.0	N/A	
Positive reaction range	N/A	All Negative	N/A	All Negative	3.0	N/A	
Clinical site Totals							
Total tested	1	6	13	12	99	213	
Mean positive reaction strength	All Negative	All Negative	3.9	2.6	3.0	3.3	
Positive reaction range	All Negative	All Negative	0.5 - 6.0	0.5 - 7.0	0.5 - 7.0	0.5 - 6.0	

Table 6. Analysis of samples producing discrepant results

GN broth-enriched samples

Site	Specimen Serial #	Specimen Storage	ICS EHEC Result	Premier Absorbance	Premier Interp	CTA Result
3	2831	Cary-Blair 2-8 C	Negative	0.223	Positive	Negative
3	282	Unpreserved -70 C	Negative	0.289	Positive	Cytotoxin Positive
3	281	Unpreserved -70 C	Negative	0.387	Positive	Negative
3	280	Unpreserved -70 C	Negative	0.151	Positive	Negative
3	269	Unpreserved -70 C	Negative	0.535	Positive	Cytotoxin Positive
3	266	Unpreserved -70 C	Negative	0.159	Positive	Negative
3	289	Unpreserved -70 C	Negative	0.280	Positive	Negative

Mac broth-enriched samples

Site	Specimen Serial #	Specimen Storage	ICS EHEC Result	Premier Absorbance	Premier Interp	CTA Result
3	2831	Cary-Blair 2-8 C	Negative	0.314	Positive	Cytotoxin Positive
3	282	Unpreserved -70 C	Negative	0.174	Positive	Negative
3	281	Unpreserved -70 C	Negative	0.239	Positive	Cytotoxin Positive
3	280	Unpreserved -70 C	Negative	0.355	Positive	Cytotoxin Positive
3	110	Unpreserved -70 C	Negative	0.157	Positive	Negative

Legend: CTA = cytotoxin assay

Table 4 Clinical sensitivity and specificity tables

GN Broth Enrichment Cultures			MacConkey Broth Enrichment Cultures			Combined Broth Totals			
<i>ICS EHEC</i>	Premier	EHEC	<i>ICS EHEC</i>	Premier	EHEC	<i>ICS EHEC</i>	Premier	EHEC	
<i>ICS EHEC GN</i>	Positive	Negative	Total	Positive	Negative	Total	Positive	Negative	Total
Positive	54	0	54	58	0	58	112	0	112
Negative	7	279	286	5	281	286	12	560	572
Total	61	279	340	63	281	344	124	560	684
Sensitivity	54/61	88.5%	77.8% - 95.3%	58/63	92.1%	82.4% - 97.4%	112/124	90.3%	83.7% - 94.9%
Specificity	279/279	100.0%	98.7% - 100%	281/281	100.0%	98.7% - 100%	560/560	100.0%	99.3% - 100%
PPV	54/54	100.0%	93.4% - 100%	58/58	100.0%	93.8% - 100%	112/112	100.0%	96.8% - 100%
NPV	279/286	97.6%	95.0% - 99.0%	281/286	98.3%	96.0% - 99.4%	560/572	97.9%	96.4% - 98.9%
Correlation	333/340	97.9%	95.8% - 99.2%	339/344	98.5%	96.6% - 99.5%	672/684	98.2%	97.0% - 99.1%

Table 5 Test Data following resolution of discrepant samples by cytotoxin assay

GN Broth Enrichment Cultures			MacConkey Broth Enrichment Cultures			Combined Broth Totals			
<i>ICS EHEC</i>	Premier	EHEC	<i>ICS EHEC</i>	Premier	EHEC	<i>ICS EHEC</i>	Premier	EHEC	
<i>ICS EHEC GN</i>	Positive	Negative	Total	Positive	Negative	Total	Positive	Negative	Total
Positive	54	0	54	58	0	58	112	0	112
Negative	2	284	286	4	282	286	6	566	572
Total	56	284	340	62	282	344	118	566	684
Sensitivity	54/56	96.4%	87.7% - 99.6%	58/62	93.5%	84.3% - 98.2%	112/118	94.9%	89.3% - 98.1%
Specificity	284/284	100.0%	98.7% - 100%	282/282	100.0%	98.7% - 100%	566/566	100.0%	99.4% - 100%
PPV	54/54	100.0%	93.4% - 100%	58/58	100.0%	93.8% - 100%	112/112	100.0%	96.8% - 100%
NPV	284/286	99.3%	97.5% - 99.9%	282/286	98.6%	96.5% - 99.6%	566/572	99.0%	97.7% - 99.6%
Correlation	338/340	99.4%	97.9% - 99.9%	340/344	98.8%	97.1% - 99.7%	678/684	99.1%	98.1% - 99.7%

REPRODUCIBILITY

Assay precision, intra-assay variability and inter-assay variability were assessed with a reference panel prepared from broths inoculated with ST1 and ST2. Of the 11 samples in the reproducibility panel, 2 were prepared as high positive (HP) samples, four as low positive (LP) samples near the assay limit of detection, 4 as high negative (HN) samples just below the assay limit of detection, and one as a low negative sample (LN). Each clinical site tested the panel twice per day for three consecutive days. The expected results were obtained at each test interval at each site resulting in an assay precision of 100% with no variability. (See Table 7.)

Table 7 Results of reproducibility evaluations

Comparison to predicate

The following table compares the outcomes of trials using ImmunoCard STAT! EHEC to the data published for the predicate device, Premier EHEC.

Table 8 Comparison of the clinical performance of ImmunoCard STAT! to Premier EHEC

Performance Characteristics in Direct Comparison to Clinical Status or Condition (Combined Samples)	ImmunoCard STAT! EHEC Combined broths	Premier EHEC (predicate) Combined broths
Clinical Sensitivity	90.3%	100%
Clinical Specificity	100%	97.9%
Predictive Value of a Positive Test	100%	Not done
Predictive Value of a Negative Test	97.9%	Not done
Correlation	98.2%	98.1%
Performance characteristics		
Precision/Reproducibility	100%	100%
Linearity/reportable range	N/A	N/A
Limit of detection	ST1 = 1.25 ng/mL ST2 = 1.25 ng/mL	ST1 = 7 pg/well ST2 = 15 pg/well
Assay cutoff	N/A	OD 0.150 (dual wavelength)

CONCLUSIONS

ImmunoCard STAT! EHEC can be used to detect Shiga-toxin forming strains of *E. coli* in human stool samples. The performance of this device for the stool broth intended use is substantially equivalent to the predicate Premier EHEC



DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
2098 Gaither Road
Rockville MD 20850

Ms. Susan Rolih
Official Correspondent
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Cincinnati, OH 45244

FEB 14 2007

Re: k062546

Trade/Device Name: ImmunoCardSTAT! EHEC
Regulation Number: 21 CFR 866.3255
Regulation Name: Escherichia coli serological reagents
Regulatory Class: Class I
Product Code: GMZ
Dated: February 6, 2007
Received: February 6, 2007

Dear Ms. Rolih:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

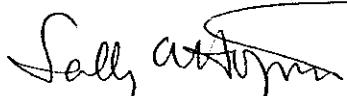
If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820).

This letter will allow you to begin marketing your device as described in your Section 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific information about the application of labeling requirements to your device, or questions on the promotion and advertising of your device, please contact the Office of In Vitro Diagnostic Device Evaluation and Safety at (240)276-0450. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21CFR Part 807.97). Other general information on your responsibilities under the Act may be obtained from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (301) 443-6597 or at its Internet address <http://www.fda.gov/cdrh/dsma/dsmamain.html>.

Sincerely yours,



Sally A. Hojvat, M.Sc., Ph.D.
Director
Division of Microbiology Devices
Office of *In Vitro* Diagnostic Device
Evaluation and Safety
Center for Devices and
Radiological Health

Enclosure

510(k) Notification
Meridian Bioscience, Inc.

ImmunoCard STAT! EHEC

INDICATIONS FOR USE STATEMENT
ImmunoCard STAT! EHEC

510(K) Number: K062546

ImmunoCard STAT! EHEC is an immunochromatographic rapid test for the qualitative detection of Shiga toxins 1 and 2 (also called Verotoxins) produced by *E. coli* in cultures derived from clinical stool specimens. ImmunoCard STAT! EHEC is used in conjunction with the patient's clinical symptoms and other laboratory tests to aid in the diagnosis of diseases caused by enterohemorrhagic *E. coli* (EHEC) infections.

Prescription Use X _____ AND/OR Over-The-Counter Use _____
(Part 21 CFR 801 Subpart D) (21 CFR 807 Subpart C)

Concurrence of CDRH, Office of In Vitro Diagnostic Devices (OIVD)

Fredrik H. Poole
Division Sign-Off

Office of In Vitro Diagnostic Device
Evaluation and Safety

510(k) K062546